OBSERVATIONS ON THE FEEDING, REPRODUCTION AND FAECES OBTAINED FROM ORIBATIDS FED ON DIFFERENT SPECIES OF *PENICILLIUM* AND *ASPERGILLUS*

G. M. SHEREEF

Department of Entomology,
Faculty of Biology, Moscow State University (U. S. S. R.)

The present paper reports some observations on some species of Oribatei found in the U. S. S. R. Complete life cycles of five species bred entirely on a diet of fungi were worked out. Besides this, feeding habit of four species was also studied.

METHODS

The first investigator, who succeeded in rearing oribatids was MICHAEL (1883-1804). He used a glass ring on a slide with a cover. SENGBUSCH (1954), PAULY (1956) and HARTENSTEIN (1962) used STENDER dishes with filter paper on the bottom. LIPOVSKY *et al.* (1957) were the first to use a mixture of plaster of Paris and charcoal.

In this investigation, all species enlisted in table 1, were collected by the author from the soil by using a Tullgren apparatus. After collecting a sufficient number of mites, the species were hand sorted. Individuals representing each species were kept alive in the laboratory in a smooth Petri dish (5 × 5 × 2 cm), which contained moistened plaster of Paris and charcoal mixed in the ratio 9 : 1. All species of mites observed were capable of living for long periods in such containers. The fungi were fed to the oribatids by providing each culture vessel with a section of agar containing an adherent portion of fungus favourable to the species. These dishes were kept in an incubator at a temperature of 25°C. The charcoal/plaster of Paris mixture was kept damp by spraying water daily. The moistened plaster of Paris kept the atmosphere in the culture vessel at approximately 100 p. 100 relative humidity and the charcoal present caused a readily visible colour change when the cultures were in danger of drying out, and also facilitated observation of the eggs of mites and spermatophores.

SENGBUSCH (1954) and LUXTON (1966) reported that mites can escape, if the vessel is not deep. In my opinion favourable diet is more important than vessel depth. Failure to culture some oribatid mites such as *Amaerus polonicus*, *Carabodes coriaceus* and *Phthiracarus* sp. that I met with was due to the lack of favourable diet. In these cases, the animals escaped from container in spite of the fact that the container was deep enough. On the other hand, sometimes in the breeding experiments the slide cover fell down, but the mites did not escape.

Dishes were examined daily under a binocular dissecting microscope using a magnification of 32 or 56 times. As soon as the eggs were discovered they were transferred, by means of small camel hair brush, to a glass ring (1 × 1 × 0.5 cm) on a slide with another slide as a cover. The

slides were held together with a rubber band. A mixture of plaster of Paris and charcoal about a quarter deep was laid at the bottom. This method helps observation of the different stages and in counting eggs and spermatophores.

Species *Belba rossica, Metabelba rara, Porobelba spinosa, Belba meridionalis,* and *Epidamaeus kamaensis* were reared on the fungus *Penicillium martensii* and the species *Oppia concolor* and *Granuloppia* sp. and *Oppia nova,* were reared on the fungus *Aspergillus flavus.*

For the feeding experiments, I used 10 species of soil fungi for *Epidamaeus kamaensis* and 8 other species for *Oppia concolor* to observe the feeding habit and the rate of multiplication of each mite on each species of fungus.

Pure cultures of fungi were obtained from the Department of Soil Biology, Moscow State University. Each species of fungus was grown axenically on wort agar in covered Petri dishes (10 × 10 × 2 cm). Petri dishes with fungi were kept in incubator at 25°C, until a thick matrix of mycelia and spores had grown over the surface of the agar slants, which usually took about three weeks.

300 tritonymphs each of *Epidamaeus kamaensis* and *Oppia concolor* were separated from a culture. After 9 days, these developed to adult stage. After another four days, with the onset of spermatophore deposition, males and females could be recognized.

Seven adults (2 males and five females) were put in a glass ring (2 × 2 × 1 cm), with a mixture of plaster of Paris and charcoal. After one month from the beginning of fungus culture, a small amount of fungus without agar was put in every glass ring. These rings were kept at 25°C and 100 p. 100 relative humidity. Each feeding experiment was repeated four times.

Rings were examined daily to determine the amount of feeding and production of faecal pellets, were provided fresh fungi when needed.

The rate of multiplication of the mites was determined by counting eggs, larvae, nymphs and adults in every ring after 104 days (from 19-12-1969 to 2-4-1970).

A fungus considered satisfactory for feeding and reproduction is denoted by plus sign (tables 3 and 4).

TABLE I

Species of oribatids found in U. S. S. R.

Species	Family	Locality
1. *Belba rossica*	Belbidae	From Caucasus
2. *Belba meridionalis*	Belbidae	From Middle-Asia
3. *Metabelba rara*	Belbidae	From Caucasus
4. *Porobelba spinosa*	Beldodamaeidae	—
5. *Epidamaeus kamaensis*	Dameidae	From Siberian
6. *Spatiodamaeus subverticillipes* ..	Damaeidae	—
7. *Oppia nova*	Oppiidae	From Moscow
8. *O. concolor*	Oppiidae	From Caucasus
9. *Granuloppia* sp.....	Oppiidae	—

RESULTS

Observation on the life-cycles

Eggs, as soon as discovered, were transferred carefully to a glass ring, and observations were continued until adult stage was reached at 25°C and 100 p. 100 relative humidity. All stages of *Belba rossica, Metabelba rare, Porobelba spinosa,* and *Epidamaeus kamaensis* were bred on the fungus *Penicillium martensii,* while *Oppia concolor* was bred on the fungus *Aspergillus flavus.*

Life cycles of these species are illustrated in table 2 (fig. 1). From this table, it is seen that life cycle of *B. rossica* is lengthier than that of other species. Moreover, it is found that in all these species, the life cycle of the males is shorter than that of the females.

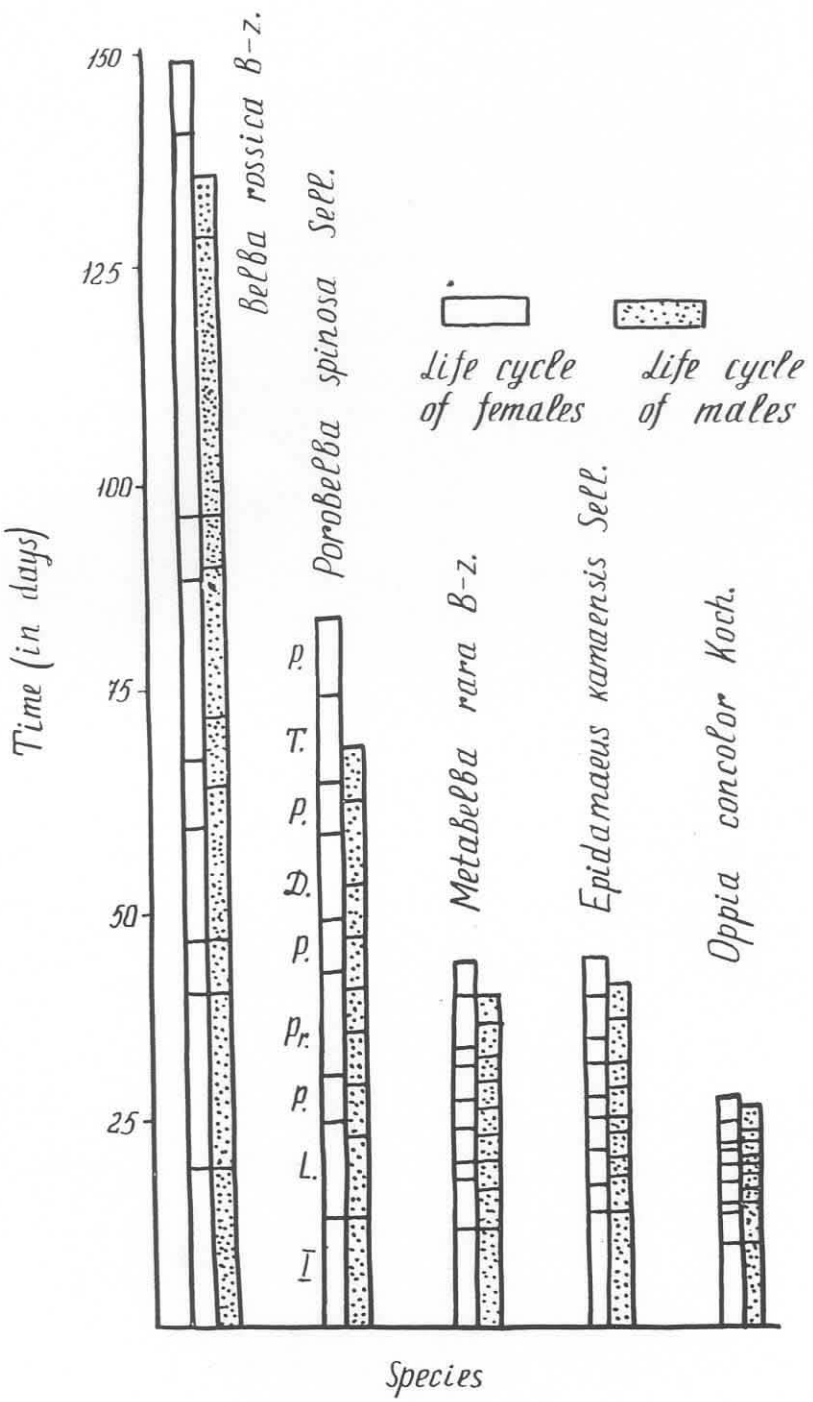

FIG. 1. — *Life cycle of 5 species of Oribatids*
Cycle de 5 espèces d'oribates

I	incubation period	période d'incubation
Pr	protonymph	protonymphe
P	pre-tritonymph	pré-tritonymphe
L	larva	larve
P	pre-protonymph	pré-protonymphe
P	pre-deutonymphe	pré-deutonymphe
D	deutonymph	deutonymphe
T	tritonymph	tritonymphe
P	pre-adult	pré-adulte

TABLE 2

Duration (in days) *of stages in the life cycles of 5 species of Oribatid mites at 25°C, and 100 p. 100 R. H.* (means based on 12 individuals)

Species	Females										Males								
	Egg	Larva	Pre-protonymph	Protonymph	Pre-deutonymph	Deutonymph	Pre-tritonymph	Tritonymph	Pre-adult	Total	Larva	Pre-protonymph	Protonymph	Pre-deutonymph	Deutonymph	Pre-tritonymph	Tritonymph	Pre-adult	Total
Belba rossica	20	20	6	14	18	21	7	44	8	148	20	6	19	8	17	6	32	7	135
Metabelba rara	12	6	2	4	3	4	2	6	4	43	5	3	3	3	3	3	4	3	39
Porobelba spinosa	14	11	5	12	6	10	6	10	9	83	9	6	6	5	6	6	10	6	68
Epid. kamaensis	14	3	4	4	2	4	3	5	4	43	4	2	3	2	3	3	5	4	40
Oppia concolor	10	4	1	2	2	2	1	2	3	27	5	1	2	1	1	2	1	3	26

According to MICHAEL (1883), the time from egg deposition to adult emergence for *Oppia nitens* was 40 days, for *Ceratoppia bipilis* 79 days and for *Cepheus palmicinctum* 375 days. PAULY (1956) determined the life cycle and total eggs for *Belba clavipes* 75 days, 60 eggs, *B. gracilipes* 75 days, 70 eggs and for *B. geniculosa* 150 days and 40 eggs. Length of the life cycle of *Spatiodamaeus boreus* was 120 days according to SITNIKOVA (1959). WOODRING and COOK (1962) reported that the life cycles of *Ceratozetes cisalpinus, Scheloribates laevigatus* and *Oppia neerlandica* were completed in 32 days, 64 days and 23 days respectively and the total number of eggs which were produced by these species were 10-16, 12 and 12 respectively.

SENGBUSCH (1954) was the first investigator, who gave the percentage of survival in his cultures of three species of Galumnids. He reported that 42 p. 100 survived, which he considered excellent.

In this investigation, it was observed that the percentage of survival in breeding experiments were 90 p. 100 in *Oppia concolor*, 85 p. 100 in *Belba rossica*, 73 p. 100 in *Epidamaeus kamaensis* 53 p. 100 in *Porobelba* spinosa and 38 p. 100 in *Metabelbarara.*

HARTENSTEIN (1962) reported that development of *B. kingi* was most rapid for mites whose only food consisted of fungus *Trichoderma koningii* or those whose culture also included *Cladosporium cladosporoides* and a *Stemphylium* sp. Survival was very low when particles of decaying leaves were added to a culture containing *B. kingi* immatures and *T. koningii*. On the other hand, WOODRING and COOK (1962), reported that when *Ceratozetes cisalpinus* was reared on the fungal hyphae alone, only 10 p. 100 onthe eggs that hatch succeeded to complete their life cycle to adult stage. Even these adults were short lived, and never produced any eggs.

My observations show that a fungus diet is very important to mites for completing their life cycles. In case of *B. rossica* and *Spatiodamaeus subverticillipes*, the author tried to breed these species on clean hay, but all larvae (22) died. It was further observed that, when the mites were put with natural food (rotten wood, moss, green grass) and fungi in Petri dish, after some time (about 2 hours) 95 p. 100 of the mites were found on the fungus. Oribatid mites were successfully cultured on the fungi *Penicillium* and *Aspergillus* alone from November 1968 till now.

Moreover, when one male and one female were put together in a glass ring, the female began to deposite eggs after 110 days in *B. rossica*, 14 days in *P. spinosa*, 10 days in *E. kamaensis* and 5 days in *O. concolor*. Total number of eggs produced by *E. kamaensis* was 84 on the average, 17 by *P. spinosa* and 23 eggs by *O. concolor.*

Life span, at 25°C and 100 p. 100 R. H., varies greatly from species to species. Individual adults of *B. rossica* lived 326 days, but *Metabelba rara* lived only 122 days. *Oppia concolor* lived 6 months and 19 days. *E. kamaensis* lived 193 days and *P. spinosa* 5 months.

In these species copulation was never observed. All these species could produce spermatophores.

Feeding and reproduction of E. kamaensis *and* Oppia concolor *on different species of fungi*

SINHA and MILLS (1968) experimented with 10 different species of *Penicillium* and concluded that *P. cyclopium* was the most favourable and *P. implicatum* the least favourable diet for *Acarus siro* and *Tyrophagus putrescentiae*. According to LUXTON

(1966), who cultured three species of oribatid mite on 10 species of fungi, the adult mites showed food preference for *Cercospora solanae*, while the larvae and nymphs were attracted to *Fusarium solani*. In an experiment on soil algae as a food source for 8 species of oribatid mites, three (*Porobelba spinosa*, *Adoristes ovatus* and *Odontoce-*

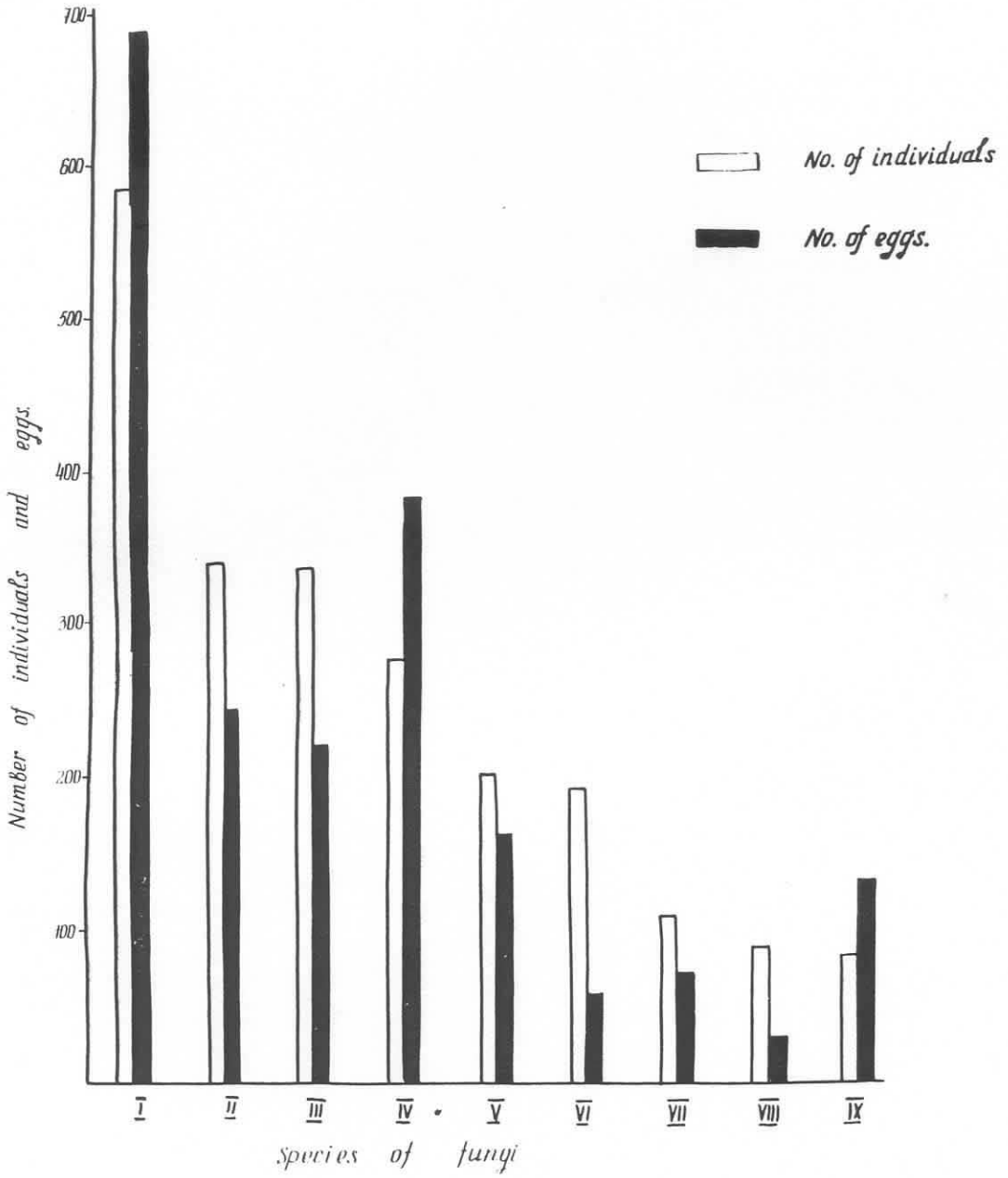

FIG. 2. — *Egg laying and reproduction of* Epidamaeus kamaensis *in 104 days on 9 different species of fungi*
*Ponte et reproduction d'*Epidamaeus kanaensis *en 104 jours sur 9 espèces différentes de champignons*

I *Trichoderma* sp.
II *Mucor ramannianus*
III *Fusarium* sp.
IV *Penicillium spinulasum*
V *P. brevi-compactum*
VI *Penicillium viridicatum*
VII *P. martensii*
VIII *P. cyclopium*
IX *P. lilacnium*

pheus elongatus) could not produce eggs while the rest, *Camisia segnis*, *C. spinifer*, *Platynothrus peltifer*, *Ceratoppia bipilis* and *Damaeus onustus* deposited eggs and larvae developed (LITTLEWOOD, 1966).

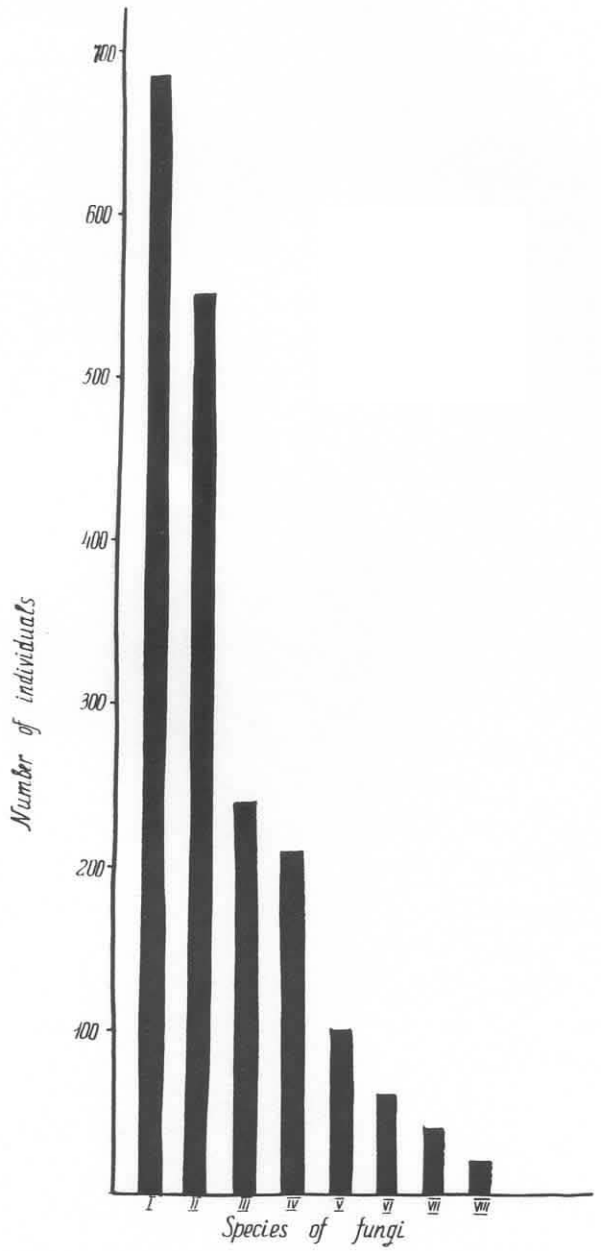

FIG. 3. — *Reproduction of* Oppia concolor *in 104 days on 8 species of fungi*
Reproduction de O. concolor *en 104 jours sur 8 espèces de champignons*

I *Fusarium* sp.
II *Aspergillus flavus*
III *A. tamerii*
IV *A. versicolor*
V *A. niger*
VI *A. flavipes*
VII *A. ustus*
VIII *A. ochraceus*

In the present investigations, soil fungi were used as food for two species of oribatid mites. Five virgin females and two males were put in every glass ring (2 × 2 × 1 cm) containing fungi. *Epidamaeus kamaensis* was served with 10 different species of fungi ;

and *Oppia concolor* with 8 species of *Aspergillus* and one species of *Fusarium* sp.

From table 3 and figure 2, it is seen that *Trichoderma* sp. was the most favourable diet for *E. kamaensis*. *Penicillium spinulasum* looks like the most attractive of the *Penicillium* species.

In case of *Oppia concolor*, it was observed that *Fusarium* sp. was very suitable. On the other hand, *Aspergillus flavus* was the most and *A. fumigatus* the least favourable diet for this species of mite (table 4 and fig. 3). None of these mites could reproduce on agar alone.

The results presented in tables 3 and 4 and figures. 2 and 3 show that both species are fungivores. Moreover, it is known that continued reproduction of a species of mite when fed on a species of fungus is a measure of the nutritional adequacy of that fungus (SINHA and WHITNEY, 1969).

The number of eggs produced by *Epidamaeus kamaensis* varies greatly according to the source of food. Larger numbers of eggs were observed on the fungus *Trichoderma* sp. (695), than on *Penicillium cyclopium* (15).

The rate of multiplication of *E. kamaensis* and *Oppia concolor* fed on different species of fungi is shown in tables 3 and 4 respectively.

Quantity of fungi consumed by mites

In the literature, no data about the food quantity consumed by oribatid mites in nature or in the laboratory was available.

300 adults of *E. kamaensis* were supplied with 0.05 g of *Penicillium martensii* without agar. The mites consumed the entire quantity of fungus in 4.8 days on the average. Equal numbers of *Belba meridionalis* took 5.5 days (average) to consume the same amount of food while only 92 individuals of *Spatiodamaeus subverticillipes* finished a similar quantity of fungus in 4 days.

300 adults of *Oppia nova* and *Granuloppia* sp. consumed 0.05 g of the fungus *Aspergillus flavus* in 19 and 11 days (average) respectively.

BYZOVA (1964) reported that the total number of oribatid mites, in forest soils of the U.S.S.R, exceeded 245 920 individuals /m². Also fungi are abundant in soils, about 3 984.000 fungus colonies originated per g of organic matter in a hemlock forest according to COBB (1932).

From these data and the above facts, oribatid mites can consume large quantity of fungi every day (approximately 82 grams of *Penicillium martensii* and 23 grams of *Aspergillus flavus*). Also, it can be concluded that soil fungi ase a great source of food for oribatid mites.

Faecal pellets

In spite of the fact that the mites habitually feed on fungal spores, they did not digest all spores. Faecal pellets, as soon as they were obtained from mites, were put in tubes containing agar. Fungal growth was observed after 24 hours. The experiment was repeated with all species which fed on *Penicillium martensii*. In every case, the result was positive. Further, mites (300 adults and nymphs of *Epidamaeus kamaensis*) which fed on *P. martensii* were washed in alcohol (90 p. 100) and formalin (40 p. 100)

and then crushed and put in a tube containing agar. Fungi were found to grow after 3 days. This was also found in case of *Oppia nova* and *O. concolor* which fed on *Aspergillus flavus*.

TABLE 3

Feeding, egg laying and reproduction of Epidamaeus kamaensis *on 10 different species of fungi*

Species	Feeding	Egg laying	No. of indiv.	No. of eggs	Tot. num.	Rate of multiplication
Trichoderma sp.	++++	++++	588	695	1 283	183
Penicillium spinulasum	+++	+++	279	385	664	95
Mucor ramannianus	+++	++	341	247	588	84
Fusarium sp.	+++	++	339	223	562	80
P. brevi-compactum	++	+	202	163	365	52
P. viridicatum	+	F	195	59	254	36
P. lilacium	+	+	85	133	218	31
P. martensii	+	F	110	73	188	27
P. cyclopium	+	F	95	15	105	15
Cladosporium sp.	—	—	—	—	—	—

In tables 3 and 4 numbers of (+) signs denote degrees of feeding and egg laying. (—) sign denotes absence of feeding and egg-laying .F — few eggs laid.

TABLE 4

Feeding and reproduction of Oppia concolor *on 9 species of fungi*

Species	Feeding	No. of indiv.	Rate of multiplication
Fusarium sp.	++++	684	98
Aspergillus flavus	++++	555	79
A. tamarii	+++	233	33
A. versicolor	++	212	30
A. niger	++	97	12
A. flavipes	+	59	8
A. ustus	+	39	6
A. ochraceus	+	19	3
A. fumigatus	—	—	—

FORSSLUND (1939) analyzed the fecal pellets of freshly caught oribatids and found mostly fungal hyphae with smaller amounts of fungal spores. LUXTON 1966) by preparations of the guts of oribatid mites, found that the same contained fungal hyphae.

These facts (feeding, breeding and reproduction on fungi) prove that oribatid

mites play an important role in the distribution of fungi in the soil and are instrumental in increasing soil fertility by helping the decomposition of organic materials and fungi.

ACKNOWLEDGEMENTS

I wish to express my deep thanks to Dr E. M. BULANOVA-ZACHVATKINA, Department of Entomology, Faculty of Biology, Moscow State University, for her help in identifying the mites.

Thanks are also due to Dr T. G. MERSHENK, Department of Soil Biology, Moscow State University, who kindly provided me with the cultures of fungi.

RÉSUMÉ

OBSERVATONS SUR LA PRISE DE NOURRITURE, LA REPRODUCTION ET LES FÉCES D'ORIBATES ALIMENTÉS SUR DIFFÉRENTES ESPÈCES DE *PENICILLIUM* ET D'*ASPERGILLUS*

La présente communication expose les résultats de recherche sur l'alimentation et la reproduction de neuf espèces d'oribates de la faune de l'U. R. S. S. Le cycle biologique de cinq d'entre elles a été calculé. Deux espèces de moisissures (*Penicillium martensii* et *Aspergillus flavus*) ont été utilisées comme source de nourriture.

La consommation et le rythme de multiplication d'*Epidamaeus kamaensis* et d'*Oppia concolor* ont été déterminés, en fournissant aux acariens, différentes espèces de moisissures. Le régime le plus favorable, ainsi que le régime le moins favorable, ont été déterminés pour ces deux espèces d'acariens.

La quantité de moisissure consommée par différentes espèces d'oribates a été calculée.

Les expériences ont montré que les pelotes fécales des oribates nourris de champignons contiennent des spores vivantes.

INDEX OF SPECIES NAMES

Species of mites

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2. *Adoristes ovatus* OUDMS, 1906.
3. *Belba clavipes* (WILLMANN, 1931 ; BUITEN, 1945) = *Damaeus clavipes* HERM., 1804.
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5. *Belba geniculosa* (OUDS, 1929 ; BUITEN, 1945) = *Damaeus onustus* (KOCH, 1841, GRAND., 1943).
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7. *Belba rossica* B.-Z., 1962.
8. *Belba kingi* HARTENSTEIN, 1962.
9. *Camisia segnis* HERM., 1804.
10. *Camisia spinifer* KOCH, 1836.
11. *Ceratoppia bipilis* HERM., 1804.
12. *Ceratozetes cisalpinus* BERL., 1908.
13. *Cepheus palmicinctum* MICH., 1884.
14. *Epidamaeus kamaensis* SELL., 1925.
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17. *Oppia concolor* KOCH., 1844.
18. *Oppia nitens* KOCH, 1836.
19. *Oppia nova* (neerlandica) OUDMS., 1900.
20. *Platynothrus peltifer* KOCH, 1839.
21. *Porobelba spinosa* SELL., 1920.

22. *Scheloribates laevigatus* KOCH, 1836.
23. *Spatiodamaeus boreus* B.-Z., 1957.
24. *Spatiodamaeus subverticillipes* B.-Z., 1957.
25. *Tyrophagus putrescentiae* (SOHRANK, 1781) ; OUDMS, 1905.

Species of fungi

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2. *Aspergillus flavipes* BAIN and SART,
3. *Aspergillus fumigatus* FRESENIUS.
4. *Aspergillus niger* VAN TIEG
5. *Aspergillus ochraceus* WILHELM.
6. *Aspergillus tamarii* KITA.
7. *Aspergillus ustus* THOM and CHURCH.
8. *Aspergillus versicolor* TIRABOSCHI.
9. *Cercospora solani* SACC.
10. *Cladosporium cladosporioides* FRES.
11. *Fusarium solani* ((MART.) APP. and WR.
12. *Mucor ramannianus* MOELLER.
13. *Penicillium brevi-compactum* DIER.
14. *Penicillium cyclopium* WESTL.
15. *Penicillium lilacinum* THOM.
16. *Penicillium martensii* BIOURGE.
17. *Penicillium spinulasum* WARCUP.
18. *Penicillium viridicatum* WESTL.
19. *Trichoderma koningii* OUDEM.

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DISCUSSION

M. J. SWIFT : You mentioned a comparison of the amount of fungal material consumed by different species of mites. Can you tell us how you measured the amount of fungal material and under what conditions ?

G. SHEREEF : I measured the amount of fungal material by electric balance. I put 0.05 g

of fungus in Petri dishes and every day I examined these dishes. When only faecal pellets were seen in Petrid ishes, it is proved that mites have consumed all fungus amount. These observations were made at 25°C.

G. J. F. PUGH : Foodstuffs contaminated with *Aspergillus flavus* become very toxic to higher animals which eat them. Was there any evidence of the mites which ate *A. flavus* spores and mycelium becoming diseased ?

G. SHEREEF : Species *Oppia concolor, Granuloppia* sp. and *Oppia nova* were reared on the fungus *Aspergillus flavus*. They fed and lived in cultures from November 1968 till now without any disease. *A. flavus* was the most favourable diet for these species. I obtained from 5 ♀♀ and 2 ♂♂ which were fed on *A. flavus* 555 individuals after 104 days. I think that mites produce antibiotic in their bodies which counteracts the effect of *A. flavus*.

G. KLEE : What substrate did you use for rearing your mite cultures before you fed them the fungi ?

What percentage of plaster to charcoal ?

G. SHEREEF: The mites were kept alive in the laboratory in a smooth Petri-dish (5 × 5 × 2 cm), which contained moistened plaster of Paris and charcoal mixed in the ratio 9 : 1.